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(54) **MULTI-COMPONENT SIMULTANEOUS MEASURING METHOD**

(57) Abstract:

PURPOSE: To simultaneously calculate the concns. of a plurality of substances dissolved in an aq. soln. only by performing the measurement of the aq. soln. by an infrared attenuation total reflection method (infrared ATR method) by preliminarily calculating absorptivity at every component in an arbitrary wave number by an infrared ATR method.

CONSTITUTION: An absorption wave number peculiar to a substance can be calculated by performing the absorption spectrum analaysis of infrared absorptiometric analysis. A wave number region of 900-1200cm^{-1} is an absorption wave common to saccharides, alcohol or org. acids and different from the absorption wave number of water as the fingerprint region of a substance. The separation of the absorption wave number of water and that of a dissolved substance becomes easy by the use of an infrared ATR method and the taking-out of the absorption of the fingerprint region becomes possible and the quantitative determination of the substance becomes possible. Therefore, a primary formula measuring the substance in a soln. and the relation between absorbancy and concn. in a specific wave number is preliminarily calculated to calculate the absorptivity of the wave number and plural simultaneous equations are solved to calculate the concns. of a plurality of substances dissolved in an aq. soln.

DETAILED DESCRIPTION

[Detailed Description of the Invention]
[0001]
[Industrial Application]This invention relates to the method of measuring the multicomponent concentration in solution simultaneously especially about the measuring method of the multicomponent concentration which used infrared attenuated total reflection spectroscopy (infrared ATR method).
[0002]
[Description of the Prior Art]Although an infrared spectroscopic analysis is the technique of qualitative analysis of identifying the ingredient in a sample with a mainly strange ingredient from the extinction characteristic of the ingredient unique absorption wave number in an infrared wave number field, this technique is applied also to the quantitative analysis. The principle used as the foundations is a principle of the Lambert veil shown by the following formula (I).
[0003]
[Formula 1]

$$A = \log \frac{I_0}{I} = \varepsilon c l \qquad (I)$$

As for the strength of the transmitted light at the time only of a solvent, and I, A is [an absorbance and I_0 / an absorbancy index and l of the strength of the transmitted light of the solution of the concentration c and epsilon] the thickness of a cell here.
[0004]That is, the absorbance in an absorption wave number unique for an ingredient is measured, and the constituent concentration is measured using the absorbancy index beforehand obtained from the principle of the Lambert veil. What is necessary is on the other hand, just to quantify independently like the time of one ingredient, when two or more solutes are in a sample and each ingredient has the absorption which does not lap with others. When absorption overlaps, the absorbance as the whole is expressed with the sum of the absorbance of each ingredient when concentration can disregard the interaction of

solutes greatly [so]. Therefore, in ****** containing multicomponent, the following formula (II) is materialized.
[0005]
[Formula 2]

$$\log \frac{I_0}{I} = A = \sum_i A_i = \sum_i \varepsilon_i c_i l \qquad (II)$$

[0006]For example, if an absorbance is measured with the two wave numbers a and b and it is made into A_a and A_b when the sample solution contains 1 and 2, the following simultaneous-equations (III) will be materialized. [two-ingredient]
[0007]
[Formula 3]

$$\begin{aligned} A_a &= (\varepsilon_{1a} c_1 + \varepsilon_{2a} c_2)\, l \\ A_b &= (\varepsilon_{1b} c_1 + \varepsilon_{2b} c_2)\, l \end{aligned} \qquad (III)$$

[0008]If four values of epsilon are known, this simultaneous-equations (III) can be solved and it can ask for each concentration of two ingredients. However, it is a case of the sample solution which dissolved in the organic solvent that these formulas are applied with an infrared-absorbing analysis method. In the solution which uses water as a solvent, the strong absorption by water is seen in an infrared region, and separation of absorption of a solute and water absorption is dramatically difficult. For this reason, in the case of the sample solution which multicomponent, such as two or more sugars, is dissolving in water, it is very difficult to measure that multicomponent concentration with sufficient accuracy simultaneously. A technique with which the multicomponent concentration in the solution which uses water etc. as a solvent can improve [accuracy] coincidence measurement is desired. On the other hand, in an HPLC analysis method, there was a problem in respect of pretreatment of a sample being complicated and serving as destructive inspection etc.
[0009]
[Problem(s) to be Solved]This invention persons found out the method of conducting simultaneous determination analysis, with infrared absorbance analytical method to the solution containing two or more ingredients which considered such infrared absorbance analysis and was made conventionally difficult. Therefore, this invention makes it a technical problem to provide the method of carrying out the quantitative analysis of the multicomponent, such as sugar in the method of measuring the multicomponent in a solution simultaneously with infrared absorbance analytical method, especially solution, alcohol, and organic acid.
[0010]
[Means for Solving the Problem]In infrared absorbance analysis, a substance can be asked for a specific absorption wave number by conducting the absorption-spectrum analysis. Sugar, alcohol, stretching vibration of C-OH origin [at organic acid] in 1200-1000 cm^{-1},

Inverse symmetry elasticity of C-O-C origin in 1150-1070 cm^{-1}, stretching vibration of C-O origin in 1085-1050 cm^{-1}, Unique absorption based on C-O is observed in 1250 cm^{-1} by stretching vibration of C-O origin, and 1200-1040 cm^{-1} as well as [stretching vibration of C-O, and 925 cm^{-1}] stretching vibration of a C-O skeleton, and 1040 cm^{-1}. It is such. A wave number field of 900-1200 cm^{-1} is an absorption wave number common to sugar, alcohol, and organic acid.
Calling this a fingerprint region of these substances, water absorption is a comparatively different absorption wave number.
Such a wave number field has been used for material identification as some specific fingerprint regions by a compound group. Although this was known comparatively well in infrared analysis, using this wave number field for a fixed quantity of a substance was not performed until now. However, this fingerprint region is measured and this invention persons find out carrying out a quantitative analysis for the first time.
[0011]In order to carry out a quantitative analysis except for influence of water by an infrared-absorption-analysis method, in transmission type analytical method adopted from the former, it is difficult. For this reason, separation of an absorption wave number of water absorption and a substance which is dissolving becomes easy by using an infrared ATR method. Thus, by separating water absorption, it becomes possible to take out absorption of a fingerprint region mentioned above, and a fixed quantity of a substance becomes possible. Each absorption spectrum of water and glucose solution which were measured by an infrared ATR method is shown in drawing 1 and drawing 2. A wave number field of like and 900-1200 cm^{-1} looked at by this spectrum is unique absorption of sugar, alcohol, and organic acid origin. By measuring this absorption, a quantitative analysis of two or more substances becomes possible.
[0012]This invention persons checked that an absorbance of an absorption spectrum of this field was additive property at sugar in a solution, alcohol, and organic acid. Therefore, as for an absorbance of this field, the above-mentioned formula (II) is materialized. This means that concentration of an ingredient can be measured by solving plural simultaneous equations of a number according to the number of ingredients in a solution which it is going to measure. That is, it asks for a linear expression which measured a substance in a solution, and relation between an absorbance in a specific wave number, and concentration beforehand, an absorbancy index of the wave number is calculated, and it becomes possible to ask for concentration of solution which is dissolving two or more substances by carrying out the solution of the plural simultaneous equations. Therefore, if an absorbancy index for every [by an infrared ATR method / in arbitrary wave numbers] ingredient is beforehand calculated in order to measure concentration of two or more substances in solution, it will become possible only by measuring by an infrared ATR extinction method of solution to ask for concentration of each ingredient. In an infrared ATR method, if it can be

used for measurement of solution, it is usable with any devices. For example, it is also possible by incorporating such an infrared ATR measurement device into in-line one to consider it as an on-line nondestructive analysis measuring device. Although it shows drawing 3 a typical structure, if measurement of a solution is possible for it even if an ATR absorber is a device of structures other than this, it is usable in it. Although there may be an infrared analysis apparatus by distributed type and Fourier transform type any, especially a Fourier transform type especially is preferred.

[0013]A method of carrying out the quantitative analysis of the multicomponent simultaneously by absorbance analysis using an infrared ATR device does not intend [new] not to have a report and having been announced until now. Although an example of this invention shows an example of solution, measurement of constituent concentration of a solution of various sorts of it is attained by choosing combination of a disengageable wave number with each ATR absorption wave number of a solvent and a solute. An example is shown below and this invention is explained to it still in detail.

[0014]

[Work example 1]In this example, an example which measured concentration of a mixed solution of grape sugar, sucrose, and fructose is shown. Infrared spectrometer used a 1600 type Fourier-transform-infrared-spectroscopic-analysis meter by PerkinElmer, Inc., and used a device which attached a thing made from GRASEBY SPECIAL LIMIT which attached a cell for ZnSe glass fluid measurement for this device as an ATR apparatus. This cell is thickness. A thing of 3.8 mm, 59.2 mm in length, and a 45-degree cut was used (drawing 3). A computer (SONY RCT-300) was connected with this, and IR data manager (PERKIN ELMER IRDM) was used and measured for software. Measurement was performed at temperature of 25 degreeC. Under these conditions, by an infrared ATR method, a water absorption spectrum and an absorption spectrum of the sample solution of grape sugar, sucrose, and fructose were measured, and differential-spectrum measurement was carried out to that water. This was chosen from an absorption peak of a mixed solution although 1038.2, 1062.8, and 1153.3cm^{-1} were chosen as the arbitrary numbers of measured waves. An absorbance in each wave number was plotted to volume molar concentration, and an analytical curve was prepared. A relation of an absorbance and volume molar concentration in each wave number of 1038.2 of grape sugar solution, 1062.8, and 1153.3cm^{-1} was shown in drawing 4. It was able to be considered that a relation of an absorbance and volume molar concentration in each wave number was a straight line. For this reason, a linear regression coefficient was able to be calculated. Similarly, a linear regression coefficient was calculated also about sucrose and fructose.

[0015]As a result, the regression coefficient in the wave number of 1038.2 of grape sugar, sucrose, fructose, and each sugar, 1062.8, and 1153.33cm^{-1} became as it is shown in the next table 1.

[0016]

[Table 1]

Regression coefficient . -------------------------------. Sugar 1038.2 cm^{-1}1062.8 cm^{-1}1153.3 cm^{-1}. -------------------------------. Grape sugar 0.0714 0.0483 0.0205 sucrose 0.0885 0.1090 0.0299 Fructose 0.0352 0.0736 0.0229 -------------------------------[0017]Next, the imitation fruit juice of imagination was prepared [becoming prescribed concentration using grape sugar, sucrose, and fructose, and] supposing each fruit juice component, such as nothing, a mandarin orange, an apple, and a peach. Prescribed concentration here is adjusted according to the sugar concentration of each fruit juice currently indicated by the volume "vegetables-and-fruits preservation **** (Kenpakusha, 1977)" for Kuniyasu Ogata. Thus, the result of having measured the differential spectrum of the infrared ATR spectrum in the wave number of 900-1300-cm^{-1} of peach imitation fruit juice among the adjusted imitation fruit juice was shown in drawing 5. Each imitation fruit-juice liquid showed the absorption pattern similar to the pattern shown in this figure. The absorbance in 1038.2 of this differential spectrum, 1062.8, and 1153.3cm^{-1} was shown in Table 2.

[0018]

[Table 2]

Absorbance . -------------------------------. Imitation fruit-juice . -----------------------. Absorption wave number Nothing Mandarin orange Apple Peach . -------------------------------. 1038.2cm^{-1}0.0202 0.0221 0.0278 0.0175 1062.8cm^{-1}0.0286 0.0278 0.0373 0.0229 1153.3cm^{-1}0.0096 0.0085 0.0121 0.0067. -------------------------------[0019]From the above result, the following simultaneous-equations equations 4 were able to be drawn as l= 1 based on the equation 3.

[Formula 4]

$$\begin{aligned} A_1 &= a_{g1} c_g + a_{s1} c_s + a_{f1} c_f \\ A_2 &= a_{g2} c_g + a_{s2} c_s + a_{f2} c_f \qquad \text{(IV)} \\ A_3 &= a_{g3} c_g + a_{s3} c_s + a_{f3} c_f \end{aligned}$$

Here, A is an absorbance in the wave number, a means a regression coefficient, c means volume molar concentration, and 1, 2, and 3 mean the wave number of 1038.2, 1062.8, and 1153.3cm^{-1}. g, s, and f show grape sugar, sucrose, and fructose, respectively.

[0020]By solving the above-mentioned simultaneous-equations (IV), each volume molar concentration of grape sugar of nothing, a mandarin orange, an apple, and each imitation fruit juice of a peach, sucrose, and fructose was able to be obtained. The result is shown in Table 5 from Table 3.

[0021]

[Table 3]

Grape sugar concentration . ---------------------------------. Nothing Mandarin orange Apple Peach . ---------------------------------. Measured value 0.132 by the infrared ATR method of

this invention 0.076 0.142 0.028 Concentration 0.131 prepared 0.080 0.139 0.044 -------------------------------[0022]

[Table 4]

Sucrose concentration . --------------------------------. Nothing Mandarin orange Apple Peach . ---------------------------------. Measured value 0.004 by the infrared ATR method of this invention 0.141 0.071 0.143 Concentration 0.018 prepared 0.157 0.091 0.157 ---------------------------------[0023]

[Table 5]

Fructose concentration . --------------------------------. Nothing Mandarin orange Apple Peach . ---------------------------------. Measured value 0.295 by the infrared ATR method of this invention 0.119 0.308 0.081 Concentration 0.295 prepared 0.091 0.297 0.053 ---------------------------------[0024]The above result of the preparation concentration which carried out weighing beforehand, and density measurement by the infrared ATR method of this invention corresponded very well. It became clear that the concentration of grape sugar of the imitation fruit-juice sample solution, sucrose, and fructose can be simultaneously measured with sufficient accuracy with the measuring method by this invention.

[0025]

[Work example 2]In this example, the example which measured the concentration of the commercial lactic acid bacteria beverage and the sugar component of orange juice is shown.

(1) As sugar to constitute, it is checked by the lactic acid bacteria beverage lactic acid bacteria beverage that milk sugar, grape sugar, fructose, and sucrose contain. The absorption spectrum by the infrared ATR method of a lactic acid bacteria beverage and the differential spectrum of the water absorption spectrum were shown in drawing 6. Using the same device as Example 1, about grape sugar, sucrose, fructose, and milk sugar in the density range of 0 - 2.0 mol/l. Each solution was adjusted and the analytical curve which shows the relation of the absorbance per the same molar concentration as Example 1 by an infrared ATR method with the wave number of 1033 cm^{-1}, 1053 cm^{-1}, 1063 cm^{-1}, and 1074 cm^{-1} was prepared. This analytical curve as well as Example 1 turned into a straight line, and the linear regression coefficient was calculated similarly. This regression coefficient was shown in Table 6.

[0026]

[Table 6]

A regression coefficient. ----------------------------------. Sugar 1033 cm^{-1}1053 cm^{-1}1063 cm^{-1}1074 cm^{-1}. ----------------------------------. Grape sugar 0.0764 0.0523 0.0483. 0.0575 Sucrose 0.0758 0.1200 0.1106 0.0840 Fructose 0.0283 0.0488 0.0712 0.0527 Milk sugar 0.1136 0.1083 0.1046 0.1254. ----------------------------------[0027]Subsequently, the absorption spectrum of the commercial lactic acid bacteria beverage (the tanker ace, made in SNOW BRAND Laurie, Inc.) was measured, and it asked for the absorbance. The

measurement result of the absorbance was shown in the next table 7.
[0028]
[Table 7] The absorbance of a lactic acid bacteria beverage. ---------------- absorption wave number absorbance ---------------- 1033 cm -- $^{-1}$0.04231053-cm^{-1}0.04471063-cm^{-1}0.04581074-cm^{-1}0.0420 ---------------- [0029]Based on equation (III), the following simultaneous-equations (V)s were able to be drawn as I= 1 from the regression coefficient of Table 6, and the absorbance of Table 7.
[0030]
[Formula 5]

$$
\begin{aligned}
A_1 &= a_{g1} c_g + a_{s1} c_s + a_{f1} c_f + a_{l1} c_l \\
A_2 &= a_{g2} c_g + a_{s2} c_s + a_{f2} c_f + a_{l2} c_l \qquad (V) \\
A_3 &= a_{g3} c_g + a_{s3} c_s + a_{f3} c_f + a_{l3} c_l \\
A_4 &= a_{g4} c_g + a_{s4} c_s + a_{f4} c_f + a_{l4} c_l
\end{aligned}
$$

Here, A is an absorbance in the wave number, and 1, 2, 3, and 4 mean the wave number of 1033, 1053, and 1063 or 1074 cm^{-1}. a shows an absorbancy index and, as for c, volume molar concentration, g, s, f, and l show grape sugar, sucrose, fructose, and milk sugar, respectively.
[0031]The concentration of each ingredient can be obtained by carrying out the solution of this formula about c. Thus, the measurement result of the acquired this invention method and the result of having obtained each constituent sugar with the HPLC analysis method were shown in Table 8.
[0032]
[Table 8]
Sugar concentration [of a lactic acid bacteria beverage] . --------------------------. Sugar Infrared ATR method HPLC method ------------------ -------- grape sugar 56.38 of this invention 51 sucrose 50.75 53 Fructose 30.76 44 Milk sugar 7.37 6 --------------------------
[0033]The analytical value of each ingredient was well in agreement in an HPLC analytical value.
[0034](2) It is checked by orange juice orange juice as sugar to constitute that grape sugar, fructose, and sucrose contain. We decided to analyze using the regression coefficient measured among the analytical curves measured in Example 2 (1) with the wave number of 1033 cm^{-1}, 1053 cm^{-1}, and 1063 cm^{-1} **. The absorbance measured with these three wave numbers was shown in Table 9.
[0035]
[Table 9] Absorbance --------------- absorption wave number of orange juice Absorbance --------------- 1033-cm^{-1}0.02711053-cm^{-1}0.03441063-cm^{-1}0.0414 --------------- [0036]The solution of the simultaneous equations of 3 yuan was carried out from this absorbance, and it asked

for each sugar concentration in orange juice. Sugar concentration was measured by HPLC like the above-mentioned Example 1 (1). Thus, the obtained measurement result and the result of having obtained each constituent sugar with the HPLC analysis method were shown in Table 10.

[0037]

[Table 10]

Sugar concentration [in orange juice] . --------------------------. Sugar Infrared ATR method HPLC method ------------------ -------- grape sugar 26.42 of this invention 26.5 sucrose 57.37 42.0 Fructose 22.23 26.8 --------------------------[0038]The analytical value of each ingredient by the infrared ATR method of this invention was well in agreement in an HPLC analytical value.

[0039]

[Effect of the Invention]By this invention, simultaneous [by an infrared ATR method / in a solution / multicomponent] and nondestructive analysis become possible. In particular, measurement of sugar, alcohol, organic acid, etc. is attained by using the wave number of 900-1300 cm^{-1} for measurement.

[Translation done.]

CLAIMS

[Claim(s)]
[Claim 1]To a solution containing two or more ingredients, it is the same as the number of these ingredients, or with or arbitrary numbers beyond it of infrared absorption wave numbers. Measure an absorbance by an infrared ATR method and beforehand by each ingredient independent. It asks for relation between an absorbance by an infrared ATR method in each arbitrary wave number selected, respectively, and concentration of each ingredient, A measuring method of constituent concentration of a solution containing two or more ingredients by an infrared ATR method asking for concentration of two or more ingredients simultaneously by building plural primary simultaneous equations showing relation between concentration and an absorbance using a value of an absorbance in each wave number of the above-mentioned solution, and solving these simultaneous equations.
[Claim 2]A way according to claim 1 a solution containing two or more ingredients is solution.
[Claim 3]The measuring method according to claim 1 or 2 which is a substance in which a measurement component has specific absorption in infrared absorption field 900-cm^{-1} - 1200 cm^{-1}[Claim 4]A way according to claim 3 substances with specific absorption are sugar, alcohol, and organic acid.

[Translation done.]